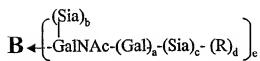
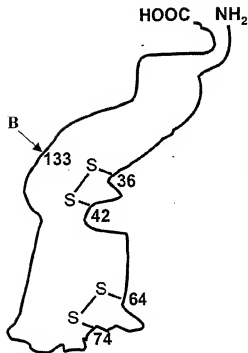


60/498



a-c, e (independently selected) = 0 or 1;

d = 0;

R = modifying group, sialyl or oligosialyl

FIG. 29A

61/498

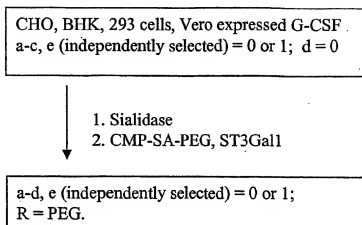


FIG. 29B

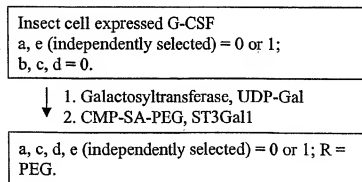


FIG. 29C

62/498

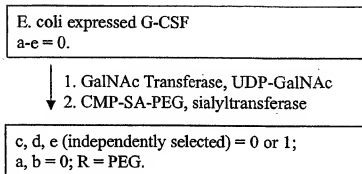


FIG. 29D

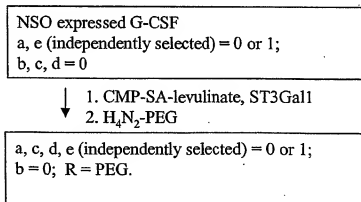


FIG. 29E

63/498

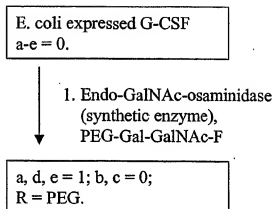


FIG. 29F

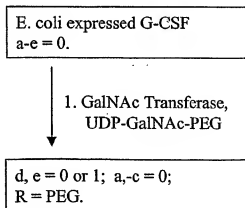
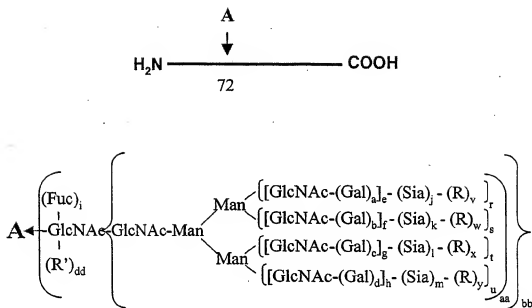


FIG. 29G

64/498



a-d, i, n-u (independently selected) = 0 or 1.

aa, bb, cc, dd, ee (independently selected) = 0 or 1.

e-h (independently selected) = 0 to 6.

j-m (independently selected) = 0 to 20.

v-z = 0; R = modifying group, mannose, oligo-mannose.

R' = H, glycosyl residue, modifying group,

glycoconjugate.

FIG. 30A

65/498

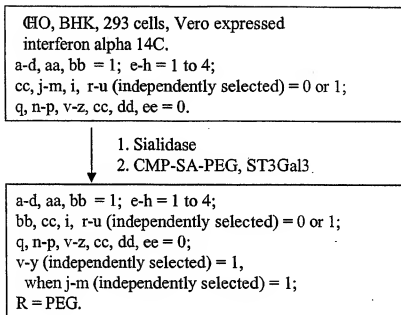


FIG. 30B

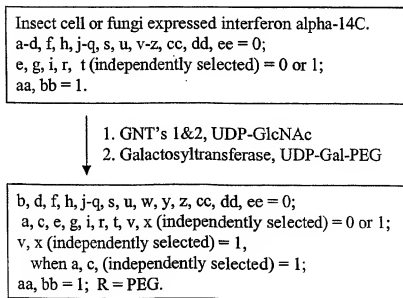


FIG. 30C

66/498

Yeast expressed interferon alpha-14C.

a-q, cc, dd, ee, v-z = 0;

r-y (independently selected) = 0 to 1;

aa, bb = 1;

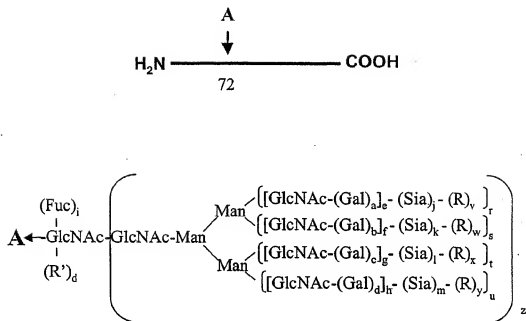
R (branched or linear) = Man, oligomannose or polysaccharide.

- ↓
1. Endo-H
 2. Galactosyltransferase, UDP-Gal
 3. CMP-SA-PEG, ST3Gal3

a-z, bb = 0; aa = 1; R' = -Gal-Sia-PEG.

FIG. 30D

67/498



a-d, i, r-u (independently selected) = 0 or 1.

e-h (independently selected) = 0 to 4.

j-m (independently selected) = 0 or 1.

n, v-y = 0; z = 0 or 1.

R = polymer; R' = sugar, glycoconjugate.

FIG. 30E

68/498

CHO, BHK, 293 cells, Vero expressed
interferon alpha-14C.

h = 1 to 3;

a-g, j-m, i (independently selected) = 0 or 1;

r-u (independently selected) = 0 or 1;

n, v-y = 0; z = 1.



1. CMP-SA-PEG, ST3Gal3

h = 1 to 3;

a-g, i (independently selected) = 0 or 1;

r-u (independently selected) = 0 or 1;

j-m, v-y (independently selected) = 0 or 1;

z = 1; n = 0; R = PEG.

FIG. 30F

Insect cell or fungi expressed
interferon alpha-14C.

a-d, f, h, j-n, s, u, v-y = 0;

e, g, i, r, t (independently selected) = 0 or 1;

z = 1.



1. GNT's 1,2,4,5, UDP-GlcNAc

2. Galactosyltransferase, UDP-Gal

3. CMP-SA-PEG, ST3Gal3

a-m, r-y (independently selected) = 0 or 1;

z = 1; n = 0; R = PEG.

FIG. 30G

69/498

Yeast expressed interferon alpha-14C.
a-n = 0; r-y (independently selected) = 0 to 1;
z = 1; R (branched or linear) = Man,
oligomannose.

- ↓
1. mannosidases
 2. GNT's 1,2,4,5, UDP-GlcNAc
 3. Galactosyltransferase, UDP-Gal
 - 4.. CMP-SA-PEG, ST3Gal3

a-m, r-y (independently selected) = 0 or 1;
z = 1; n = 0; R = PEG.

FIG. 30H

NSO expressed interferon alpha 14C.
a-i, r-u (independently selected) = 0 or 1;
j-m, n, v-y = 0; z = 1.

- ↓
1. CMP-SA-levulinate, ST3Gal3,
buffer, salt
 2. H_4N_2 -PEG

a-i, j-m, r-y (independently selected) = 0 or 1;
n = 0; z = 1; R = PEG.

FIG. 30I

70/498

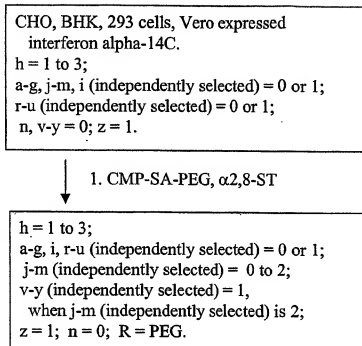


FIG. 30J

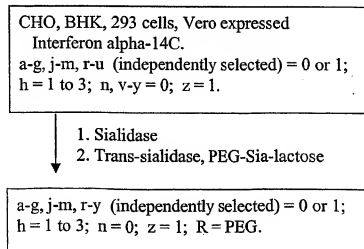


FIG. 30K

71/498

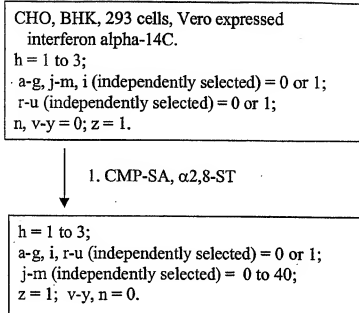


FIG. 30L

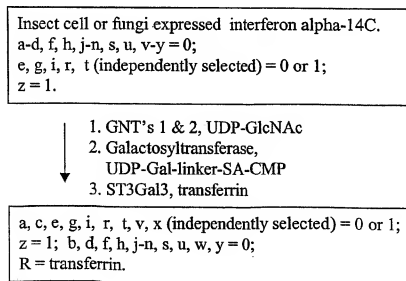


FIG. 30M

72/498

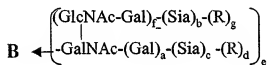
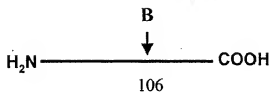
Insect cell or fungi expressed interferon alpha-14C.
a-d, f, h, j-n, s, u, v-y = 0;
e, g, i, r, t (independently selected) = 0 or 1; z = 1.

- ↓
1. endoglycanase
 2. Galactosyltransferase,
UDP-Gal-linker-SA-CMP
 3. ST3Gal3, transferrin

i (independently selected) = 0 or 1;
a-h, j-m, r-z = 0;
n = 1; R' = -Gal-linker-transferrin.

FIG. 30N

73/498



a-c, e, f (independently selected) = 0 or 1;
 d, g = 0; R = polymer, glycoconjugate.

FIG. 300

74/498

CHO, BHK, 293 cells, Vero expressed
IF-alpha (2a or 2b).
a-c (independently selected) = 0 or 1;
e = 1; d, f, g = 0

- ↓
1. Sialidase
 2. CMP-SA-PEG, ST3Gal1

a-d (independently selected) = 0 or 1;
e = 1; b, f, g = 0; R = PEG.

FIG. 30P

Insect cell expressed interferon alpha (2a or 2b).
a, e (independently selected) = 0 or 1;
b, c, d, f, g = 0.

- ↓
1. Galactosyltransferase, UDP-Gal
 2. CMP-SA-PEG, ST3Gal1

a, c, d, e (independently selected) = 0 or 1;
b, f, g = 0; R = PEG.

FIG. 30Q

75/498

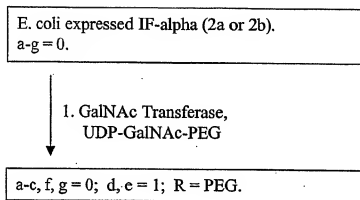


FIG. 30R

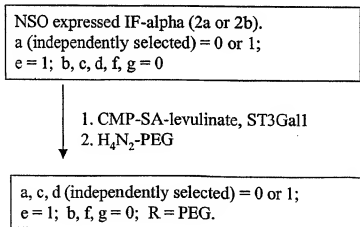


FIG. 30S

76/498

E. coli expressed IF-alpha (2a or 2b).
a-g = 0.



1. Endo-N-acetylgalactosamidase
(synthetic enzyme),
PEG-Gal-GalNAc-F

a, d, e = 1; b, c, f, g = 0; R = PEG.

FIG. 30T

E. coli expressed IF-alpha (2a or 2b).
a-g = 0.



1. GalNAc Transferase, UDP-GalNAc
2. sialyltransferase, CMP-SA-PEG

b, d = 0 or 1; e = 1; a, c, f, g = 0; R = PEG.

FIG. 30U

77/498

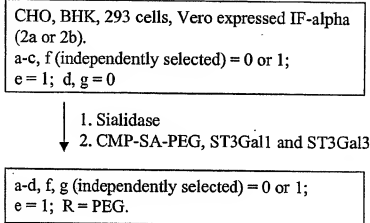


FIG. 30V

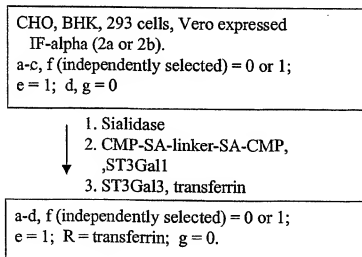
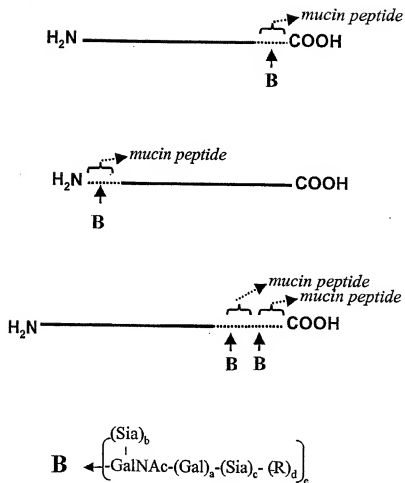


FIG. 30W

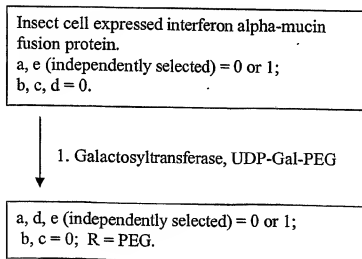
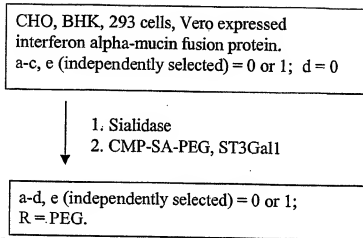
78/498



a-c, e (independently selected) = 0 or 1;
 d = 0; R = polymer, glycoconjugate.

FIG. 30X

79/498



80/498

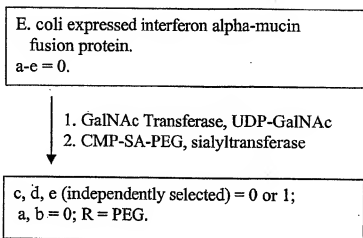
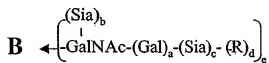
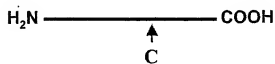
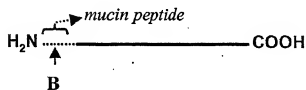
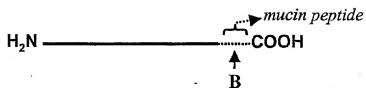


FIG. 30AA

81/498



a-c, e (independently selected) = 0 or 1;
d = 0; R = polymer, linker.

FIG. 30BB

82/498

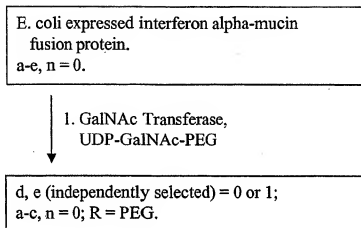


FIG. 30CC

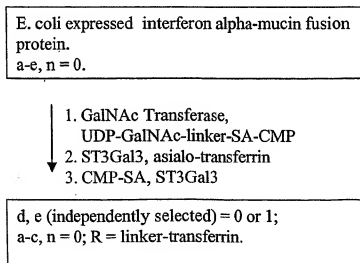


FIG. 30DD

83/498

E. coli expressed Interferon alpha (no fusion).
a-e, n = 0.

1. NHS-CO-linker-SA-CMP
2. ST3Gal3, transferrin

a-e = 0; n = 1; R' = linker-transferrin.

FIG. 30EE

85/498

CHO, BHK, 293 cells, Vero expressed IF-beta
 h = 1 to 3;
 a-g, j-m, i (independently selected) = 0 or 1;
 r-u (independently selected) = 0 or 1;
 n, v-y = 0; z = 1.

- ↓
1. Sialidase
 2. CMP-SA-PEG, ST3Gal3

h = 1 to 3;
 a-g, i (independently selected) = 0 or 1;
 r-u (independently selected) = 0 or 1;
 j-m, v-y (independently selected) = 0 or 1;
 z = 1; n = 0; R = PEG.

FIG. 31B

Insect cell expressed IF-beta
 a-d, f, h, j-n, s, u, v-y = 0;
 e, g, i, r, t (independently selected) = 0 or 1;
 z = 1.

- ↓
1. GNT's 1&2, UDP-GlcNAc
 2. Galactosyltransferase, UDP-Gal
 3. CMP-SA-PEG, ST3Gal3,
buffer, salt

b, d, f, h, k, m, n, s, u, w, y = 0;
 a, c, e, g, i, r, t (independently selected) = 0 or 1;
 j, l, v, x (independently selected) = 0 or 1;
 z = 1; R = PEG.

FIG. 31C

86/498

Yeast expressed IF-beta
 a-n = 0; z = 1;
 r-y (independently selected) = 0 to 1;
 R (branched or linear) = Man, oligomannose or
 polysaccharide.

- ↓ 1. Endo-H
 2. Galactosyltransferase, UDP-Gal
 3.. CMP-SA-PEG, ST3Gal3

a-m, r-z= 0; n = 1; R' = -Gal-Sia-PEG.

FIG. 31D

CHO, BHK, 293 cells, Vero expressed IF-beta
 h = 1 to 3;
 a-g, j-m, i (independently selected) = 0 or 1;
 r-u (independently selected) = 0 or 1;
 n, v-y = 0; z = 1.

- ↓ 1. CMP-SA-PEG, ST3Gal3

h = 1 to 3;
 a-g, i (independently selected) = 0 or 1;
 r-u (independently selected) = 0 or 1;
 j-m, v-y (independently selected) = 0 or 1;
 z = 1; n = 0; R = PEG.

FIG. 31E

87/498

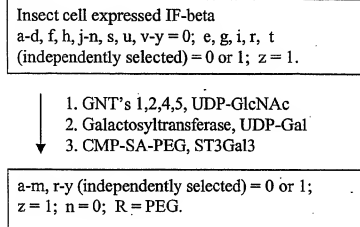


FIG. 31F

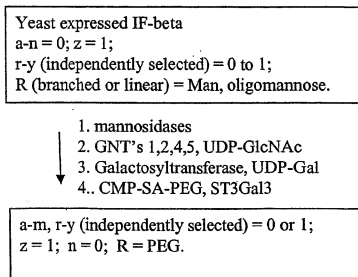


FIG. 31G

88/498

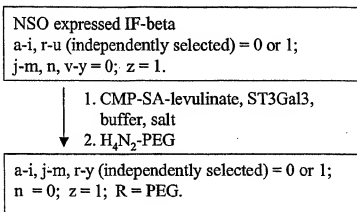


FIG. 31H

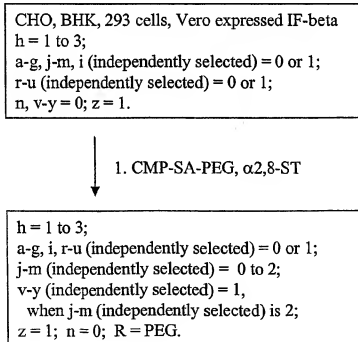


FIG. 31I

89/498

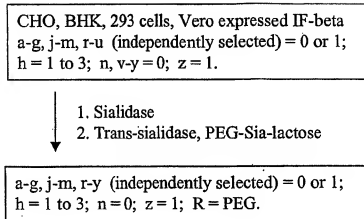


FIG. 31J

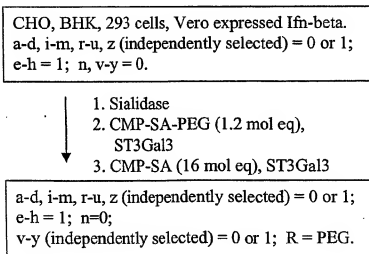


FIG. 31K

90/498

NSO expressed Ifn-beta.

a-d, i-m, r-u, z (independently selected) = 0 or 1;

e-h = 1; n, v-y = 0;

Sia (independently selected) = Sia or Gal.

- ↓
1. Sialidase and α -galactosidase
 2. α -Galactosyltransferase, UDP-Gal
 - ▼ 3. CMP-SA-PEG, ST3Gal3

a-d, i-m, r-u, z (independently selected) = 0 or 1;

e-h = 1; R = PEG

n = 0; v-y (independently selected) = 1,

when j-m (independently selected) is 1;

FIG. 31L

CHO, BHK, 293 cells, Vero expressed Ifn-beta.

a-d, i-m, r-u, z (independently selected) = 0 or 1;

e-h = 1; n, v-y = 0.

- ↓
1. Sialidase
 2. CMP-SA-PEG (16 mol eq),
ST3Gal3
 3. CMP-SA, ST3Gal3

a-d, i-m, r-u, z (independently selected) = 0 or 1;

e-h = 1; n = 0;

v-y (independently selected) = 0 or 1; R = PEG.

FIG. 31M

91/498

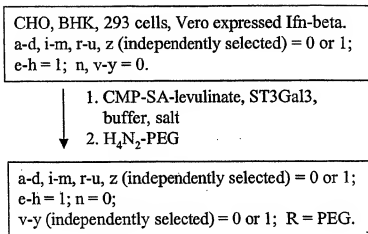


FIG. 31N

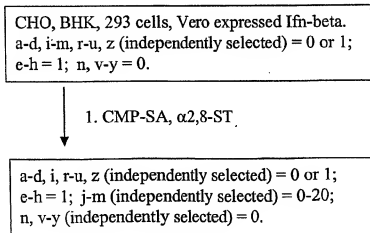
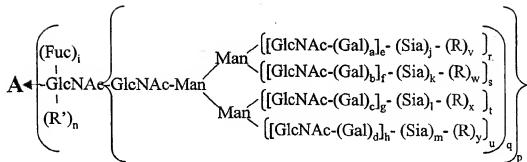
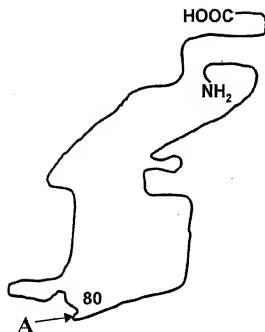


FIG. 31O

92/498



a-d, i, p-u (independently selected) = 0 or 1.

e-h (independently selected) = 0 to 6.

j-m (independently selected) = 0 to 100.

v-y = 0; R = modifying group;

R' = H, glycosyl group, modifying group,

glycoconjugate.

FIG. 31P

93/498

Insect cell expressed Ifn-beta.

a-d, f, h, j-m, s, u, v-y = 0;

e, g, i, q, r, t (independently selected) = 0 or 1.



1. GNT's 1,2,4,5, UDP-GlcNAc

2. Galactosyltransferase, UDP-Gal-PEG

a-i, q-u (independently selected) = 0 or 1;

j-m = 0; v-y (independently selected) = 1,

when e-h (independently selected) is 1;

R = PEG.

FIG. 31Q

Yeast expressed Ifn-beta.

a-m = 0; q-y (independently selected) = 0 to 1;

p = 1;

R (branched or linear) = Man, oligomannose.



1. Endoglycanase

2. Galactosyltransferase, UDP-Gal

3. CMP-SA-PEG, ST3Gal3

a-m, p-y = 0;

n (independently selected) = 0 or 1;

R' = -Gal-Sia-PEG.

FIG. 31R

94/498

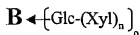
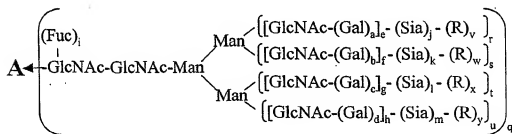
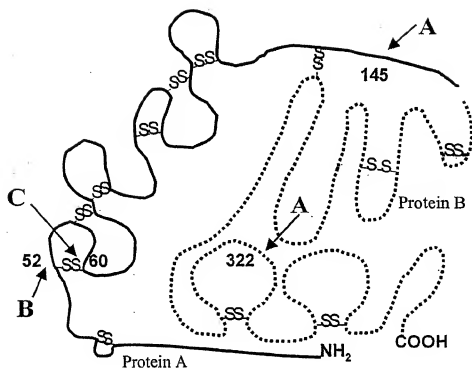
CHO, BHK, 293 cells, Vero expressed Ifn-beta.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. CMP-SA-linker-SA-CMP,
ST3Gal3
 2. ST3Gal3, desialylated transferrin.
 3. CMP-SA, ST3Gal3

a-m, q-u (independently selected) = 0 or 1;
p = 1; n = 0;
v-y (independently selected) = 0 or 1;
R = linker-transferrin.

FIG. 31S

95/498



a-d, i, q-u (independently selected) = 0 or 1.

o, p (independently selected) = 0 or 1.

e-h, n (independently selected) = 0 to 6.

j-m (independently selected) = 0 to 20.

v-y = 0;

R = modifying group, mannose, oligo-mannose, Sia-Lewis X, Sia-Lewis A..

FIG. 32A

96/498

BHK expressed Factor VII or VIIa

a-d, e, i, g, q, j, l, o, p (independently selected) = 0 or 1;
 r, t = 1; f, h, k, m, s, u, v-y = 0; n = 0-4.

- ↓
1. Sialidase
 2. CMP-SA-PEG (16 mole eq),
ST3Gal3

a-d, e, g, i, q, j, l, o, p (independently selected) = 0 or 1;
 r, t = 1; f, h, k, m, s, u, w, y = 0; n = 0-4;
 v, x, (independently selected) = 1,
 when j, l (respectively, independently selected) is 1;
 R = PEG.

FIG. 32B

CHO, BHK, 293 cells, Vero expressed Factor VII or VIIa

a-d, e, i, g, q, j, l, o, p (independently selected) = 0 or 1;
 r, t = 1; f, h, k, m, s, u, v-y = 0; n = 0-4.

- ↓
1. Sialidase
 2. CMP-SA-PEG (1.2 mole eq),
ST3Gal3
 3. CMP-SA (8 mol eq), ST3Gal3

a-d, e, g, i, q, j, l, o, p (independently selected) = 0 or 1;
 r, t = 1; f, h, k, m, s, u, w, y = 0; n = 0-4;
 v or x, (independently selected) = 1,
 when j or l, (respectively, independently selected) is 1;
 R = PEG.

FIG. 32C

97/498

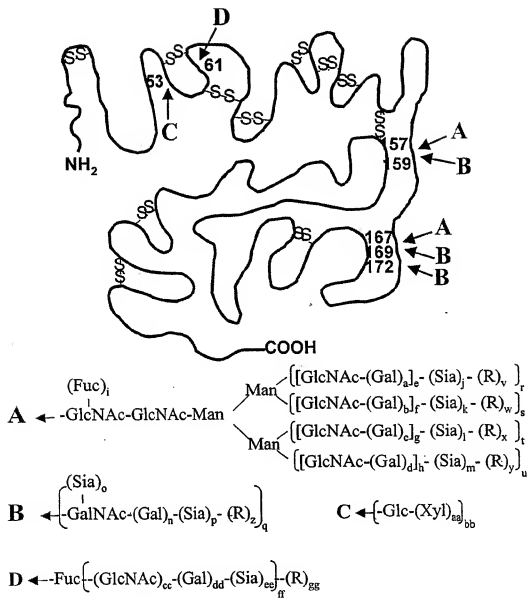
NSO expressed Factor VII or VIIa
a--u (independently selected) = 0 or 1;
v-y = 0; n = 0-4;
Sia (independently selected) = Sia or Gal.

- ↓
1. Sialidase and α -galactosidase
2. Galactosyltransferase, UDP-Gal
▼ 3. CMP-SA-PEG, ST3Gal3

a-m, o-u (independently selected) = 0 or 1;
n = 0-4; v-y (independently selected) = 1,
when j-m (independently selected) is 1;
Sia = Sia; R = PEG.

FIG. 32D

98/498



a-d, i, n-u (independently selected) = 0 or 1.

bb, cc, dd, ee, ff, gg (independently selected) = 0 or 1.

e-h, aa (independently selected) = 0 to 6.

j-m (independently selected) = 0 to 20.

v-z = 0; R = modifying group, mannose, oligo-mannose.

FIG. 33A

99/498

CHO, BHK, 293 cells, Vero expressed Factor IX
 a-d, q = 1; e-h = 1 to 4;
 aa, bb, cc, dd, ee, ff, j-m, i, n, o, p, r-u (independently
 selected) = 0 or 1;
 v-z, gg = 0.

1. Sialidase
2. CMP-SA-PEG, ST3Gal3

a-d, q = 1; e-h = 1 to 4;
 aa, bb, cc, dd, ee, ff, i, n, r-u (independently selected)
 = 0 or 1;
 o, p, z = 0;
 j-m, ee, v-y, gg (independently selected) = 0 or 1;
 R = PEG.

FIG. 33B

CHO, BHK, 293 cells, Vero expressed Factor IX
 a-d, n, q = 1; e-h = 1 to 4;
 aa, bb, cc, dd, ee, ff, j-m, i, o, p, r-u (independently
 selected) = 0 or 1;
 v-z, gg = 0.

1. Sialidase
2. CMP-SA-PEG, ST3Gal3
3. ST3Gal1, CMP-SA

a-d, n, p, q = 1; e-h = 1 to 4;
 aa, bb, cc, dd, ee, ff, i, r-u (independently selected) =
 0 or 1;
 j-m, ee, v-y, gg (independently selected) = 0 or 1;
 o, z = 0; R = PEG.

FIG. 33C

100/498

CHO, BHK, 293 cells, Vero expressed Factor IX
 a-d, n, q, bb, cc, dd, ff = 1; e-h, aa = 1 to 4; ee, j-m, i,
 o, p, r-u (independently selected) = 0 or 1; v-z, gg = 0.

- ↓
1. sialidase
 2. Galactosyltransferase, UDP-Gal
 3. CMP-SA, ST3Gal3
 4. CMP-SA-PEG, ST3Gal1

a-d, n, q = 1; e-h = 1 to 4;
 aa, bb, cc, dd, ee, ff, i, r-u (independently selected) =
 0 or 1; R = PEG;
 o, v-y, gg = 0;
 j-m, p, ee (independently selected) = 0 or 1, but when
 p = 1, z = 1.

FIG. 33D

CHO, BHK, 293 cells, Vero expressed Factor IX
 a-d, q = 1; e-h = 1 to 4;
 aa, bb, cc, dd, ee, ff, j-m, i, n, o, p, r-u (independently
 selected) = 0 or 1;
 v-z, gg = 0.

- ↓
- CMP-SA-PEG, ST3Gal3

a-d, q = 1; e-h = 1 to 4;
 aa, bb, cc, dd, ee, ff, i, n, r-u (independently selected)
 = 0 or 1; R = PEG;
 o, p, z = 0; j-m, ee, v-y, gg (independently selected) =
 0 or 1.

FIG. 33E

101/498

CHO, BHK, 293 cells, Vero expressed Factor IX
 a-d, q = 1; e-h = 1 to 4;
 aa, bb, cc, dd, ee, ff, j-m, i, n, o, p, r-u (independently
 selected) = 0 or 1;
 v-z, gg = 0.

- ↓
1. CMP-SA-levulinate, ST3Gal3,
buffer, salt
 2. H₄N₂-PEG

a-d, q = 1; e-h = 1 to 4;
 aa, bb, cc, dd, ee, ff, i, n, r-u (independently selected)
 = 0 or 1;
 o, p, z = 0; R = PEG;
 j-m, ee, v-y, gg (independently selected) = 0 or 1.

FIG. 33F

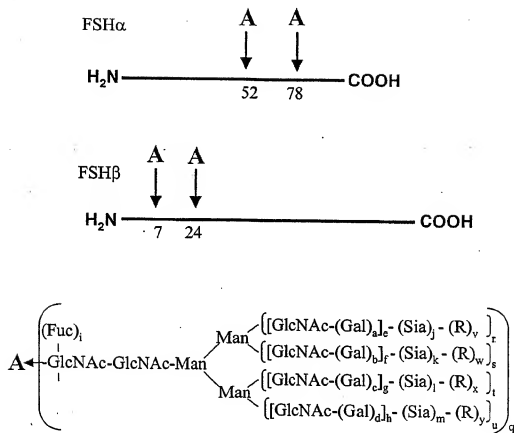
CHO, BHK, 293 cells, Vero expressed Factor IX
 a-d, n, q, bb, cc, dd, ff = 1;
 e-h, aa = 1 to 4;
 ee, j-m, i, o, p, r-u (independently selected) = 0 or 1;
 v-z, gg = 0.

- ↓
1. CMP-SA-PEG, α 2,8-ST

a-d, q = 1; e-h = 1 to 4;
 aa, bb, cc, dd, ee, ff, i, n, r-u (independently selected)
 = 0 or 1;
 o, p, z = 0; R = PEG;
 j-m, ee (independently selected) = 0 to 2;
 v-y, gg (independently selected) = 1, when j-m
 (independently selected) is 2;

FIG. 33G

102/498



a-d, i, q-u (independently selected) = 0 or 1.

e-h (independently selected) = 0 to 6.

j-m (independently selected) = 0 to 100.

v-y = 0;

R = modifying group, mannose, oligo-mannose.

FIG. 34A

103/498

CHO, BHK, 293 cells, Vero expressed FSH.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. Sialidase
 2. CMP-SA-PEG (16 mol eq),
ST3Gal3

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y (independently selected) = 1,
when j-m (independently selected) is 1;
R = PEG.

FIG. 34B

CHO, BHK, 293 cells, Vero expressed FSH.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. Sialidase
 2. CMP-SA-PEG (1.2 mol eq),
ST3Gal3
 3. CMP-SA (16 mol eq), ST3Gal3

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y (independently selected) = 0 or 1;
R = PEG.

FIG. 34C

104/498

NSO expressed FSH.

a-d, i-m, q-u (independently selected) = 0 or 1;

e-h = 1; v-y = 0;

Sia (independently selected) = Sia or Gal.

- ↓
1. Sialidase and α -galactosidase
 2. Galactosyltransferase, UDP-Gal
 - ▼ 3. CMP-SA-PEG, ST3Gal3

a-d, i-m, q-u (independently selected) = 0 or 1;

e-h = 1; v-y (independently selected) = 1,

when j-m (independently selected) is 1;

R = PEG.

FIG. 34D

CHO, BHK, 293 cells, Vero expressed FSH.

a-d, i-m, q-u (independently selected) = 0 or 1;

e-h = 1; v-y = 0.

- ↓
1. Sialidase
 2. CMP-SA-PEG (16 mol eq),
ST3Gal3
 - ▼ 3. CMP-SA, ST3Gal3

a-d, i-m, q-u (independently selected) = 0 or 1;

e-h = 1; v-y (independently selected) = 0 or 1;

R = PEG.

FIG. 34E

105/498

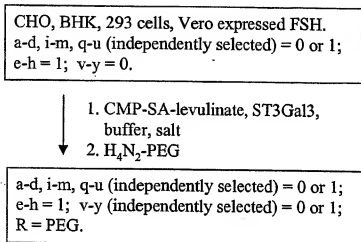


FIG. 34F

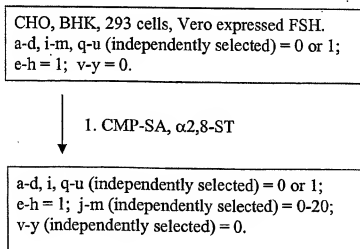


FIG. 34G

106/498

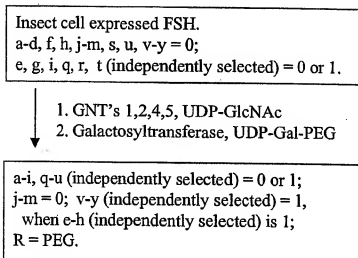


FIG. 34H

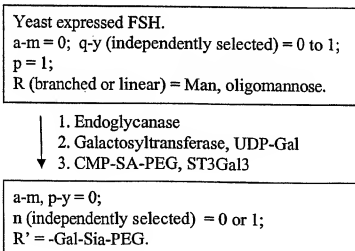


FIG. 34I

107/498

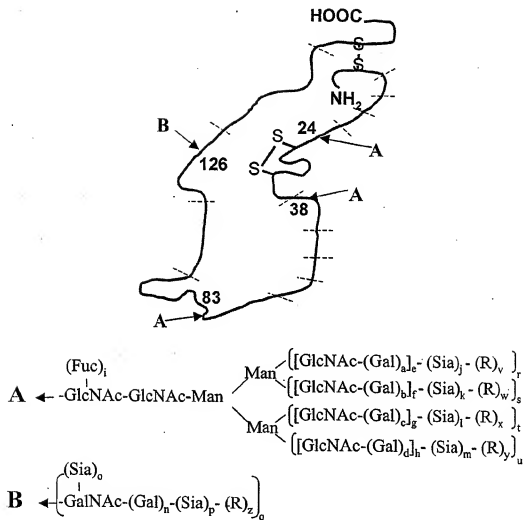
CHO, BHK, 293 cells, Vero expressed FSH.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. CMP-SA-linker-SA-CMP, ST3Gal3
 2. ST3Gal1, desialylated chorionic gonadotrophin (CG) produced in CHO.
 3. CMP-SA, ST3Gal3, ST3Gal1

a-m, q-u (independently selected) = 0 or 1;
p = 1; n = 0;
v-y (independently selected) = 0 or 1;
R = linker-CG.

FIG. 34J

108/498



a-d, i, n-u (independently selected) = 0 or 1.

e-h (independently selected) = 0 to 4.

j-m (independently selected) = 0 to 20.

v-z = 0;

R = polymer.

FIG. 35A

109/498

CHO, BHK, 293 cells, Vero expressed EPO
 a-g, n, q = 1; h = 1 to 3;
 j-m, i, o, p (independently selected) = 0 or 1;
 r-u (independently selected) = 0 to 1; v-z = 0

- ↓
1. Sialidase
 2. CMP-SA-PEG, ST3Gal3

a-g, n, q = 1; h = 1 to 3;
 i, o, p (independently selected) = 0 or 1;
 r-u (independently selected) = 0 or 1;
 j-m, v-y (independently selected) = 0 or 1;
 R = PEG; z = 0.

FIG. 35B

Insect cell expressed EPO
 a-d, f, h, j-q, s, u, v-z = 0;
 e, g, i, r, t (independently selected) = 0 or 1.

- ↓
1. GNT's 1&2, UDP-GlcNAc
 2. Galactosyltransferase, UDP-Gal
 2. CMP-SA-PEG, ST3Gal3

b, d, f, h, k, m-q, s, u, w, y, z = 0;
 a, c, e, g, i, r, t (independently selected) = 0 or 1;
 j, l, v, x (independently selected) = 0 or 1;
 R = PEG.

FIG. 35C

110/498

CHO, BHK, 293 cells, Vero expressed EPO
 a-q, r-u (independently selected) = 0 or 1;
 v-z = 0.

1. sialidase
2. Galactosyltransferase, UDP-Gal
3. CMP-SA, ST3Gal3
- ↓ 4. CMP-SA-PEG, ST3Gal1

a-h, n, q = 1;
 i-m, o, r-u (independently selected) = 0 or 1;
 v-y = 0; p, z = 0 or 1; R = PEG.

FIG. 35D

CHO, BHK, 293 cells, Vero expressed EPO
 a-g, n, q = 1; h = 1 to 3;
 j-m, i, o, p (independently selected) = 0 or 1;
 r-u (independently selected) = 0 or 1;
 v-z = 0

- ↓ 1. CMP-SA-PEG, ST3Gal3

a-g, n, q = 1; h = 1 to 3;
 i, o, p (independently selected) = 0 or 1;
 r-u (independently selected) = 0 to 1;
 j-m, v-y (independently selected) = 0 or 1;
 R = PEG; z = 0.

FIG. 35E

111/498

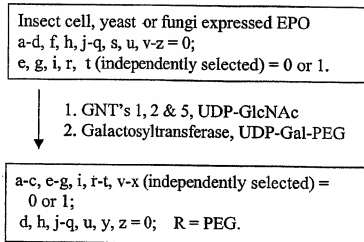


FIG. 35F

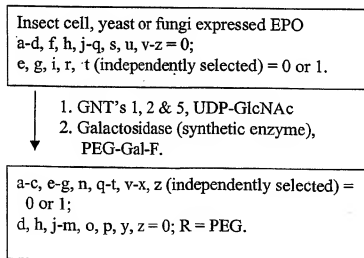


FIG. 35G

112/498

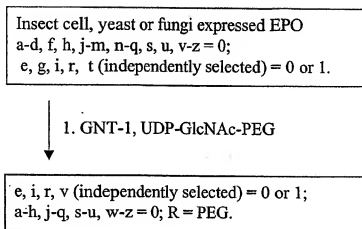


FIG. 35H

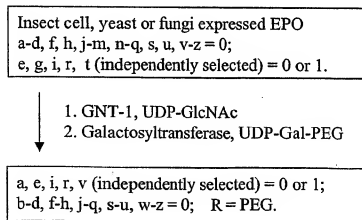


FIG. 35I

113/498

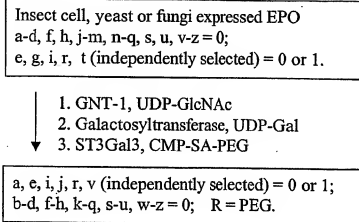


FIG. 35J

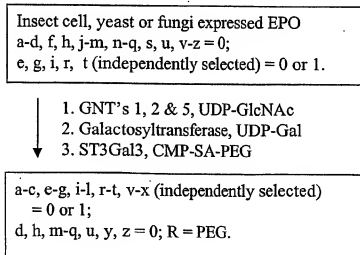


FIG. 35K

114/498

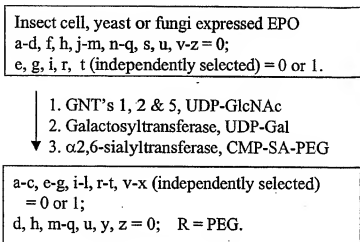


FIG. 35L

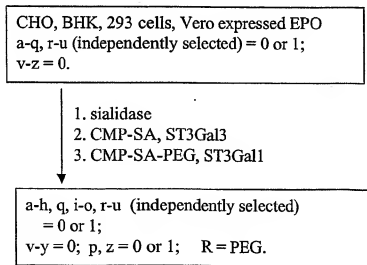


FIG. 35M

115/498

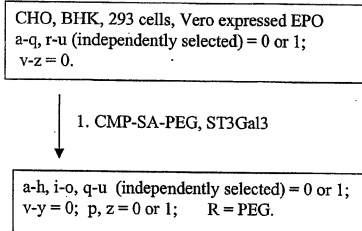


FIG. 35N

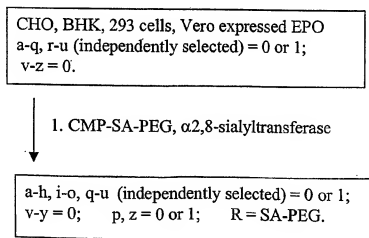


FIG. 35O

116/498

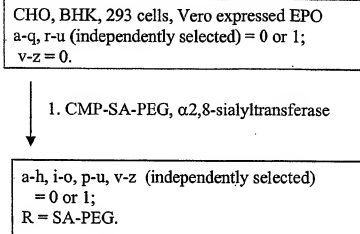


FIG. 35P

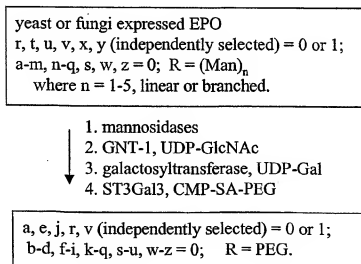


FIG. 35Q

117/498

yeast or fungi expressed EPO
 r, t, u, v, x, y (independently selected) = 0 or 1;
 a-m, n-q, s, w, z = 0; $R = (\text{Man})_n$
 where n = 1-5, linear or branched.

- ↓
1. mannosidases
 2. GNT-1, UDP-GlcNAc-PEG

c, r, v (independently selected) = 0 or 1;
 a-h, i-q, s-u, w-z = 0; $R = \text{PEG}$.

FIG. 35R

yeast or fungi expressed EPO
 r, t, u, v, x, y (independently selected) = 0 or 1;
 a-m, n-q, s, w, z = 0; $R = (\text{Man})_n$
 where n = 1-5, linear or branched.

- ↓
1. mannosidase-I
 2. GNT-1, UDP-GlcNAc
 3. galactosyltransferase, UDP-Gal
 4. ST3Gal3, CMP-SA-PEG

a, c, j, r, t-u, v, x, y (independently selected)
 = 0 or 1;
 b-d, f-i, k-q, s, w, z = 0;
 $(R)_v = \text{PEG}$; $(R)_x$ and $(R)_y = \text{Man}$.

FIG. 35S

119/498

yeast or fungi expressed EPO
 r, t, u, v, x, y (independently selected) = 0 or 1;
 cc, a-m, n-q, s, w, z = 0;
 aa, bb = 1;
 $R = (\text{Man})_n$ where $n = 1-100$, linear or branched.

- ↓
 1. endo-H
 2. galactosyltransferase, UDP-Gal-PEG

i (independently selected) = 0 or 1;
 aa, bb, cc, a-h, j-z = 0; $R'' = \text{Gal-PEG}$.

FIG. 35U

yeast or fungi expressed EPO
 r, t, u, v, x, y (independently selected) = 0 or 1;
 cc, a-m, n-q, s, w, z = 0; aa, bb = 1;
 $R = (\text{Man})_n$ where $n = 1-100$, linear or branched.

- ↓
 1. endo-H
 2. galactosyltransferase, UDP-Gal
 3. ST3Gal3, CMP-SA-PEG

i (independently selected) = 0 or 1;
 aa, bb, cc, a-h, j-z = 0; $R'' = \text{Gal-SA-PEG}$.

FIG. 35V

120/498

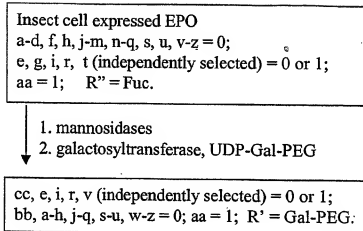


FIG. 35W

122/498

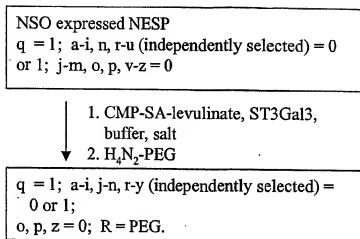


FIG. 35Y

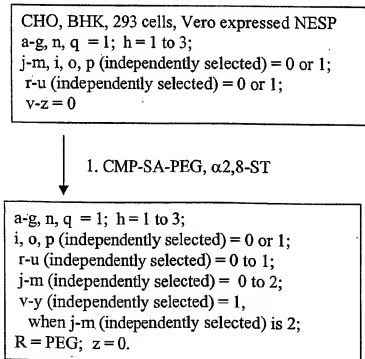


FIG. 35Z

123/498

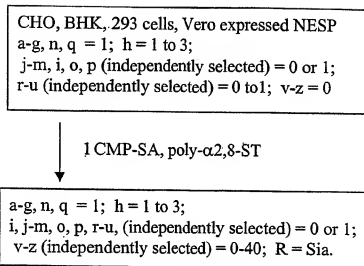
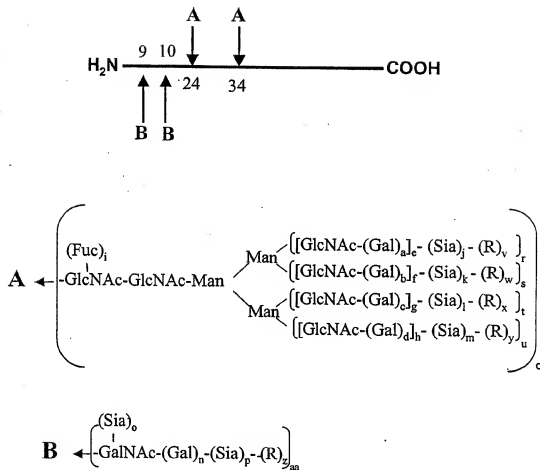


FIG. 35AA

124/498



a-d, i, n-u, aa (independently selected) = 0 or 1.

e-h (independently selected) = 0 to 6.

j-m (independently selected) = 0 to 100.

v-y = 0; R = polymer, glycoconjugate.

FIG. 36A

125/498

CHO, BHK, 293 cells, Vero expressed GM-CSF.
a-d, i-m, o-u, aa (independently selected) = 0 or 1;
n, e-h = 1; v-z = 0.

- ↓
1. Sialidase
 2. CMP-SA-PEG (16 mol eq),
ST3Gal3

a-d, i-m, q-u, aa (independently selected) = 0 or 1;
o, p, z = 0; n, e-h = 1;
v-y (independently selected) = 1,
when j-m (independently selected) is 1;
R = PEG.

FIG. 36B

CHO, BHK, 293 cells, Vero expressed GM-CSF.
a-d, i-m, o-u, aa (independently selected) = 0 or 1;
n, e-h = 1; v-z = 0.

- ↓
1. Sialidase
 2. CMP-SA-PEG (1.2 mol eq),
ST3Gal3
 3. CMP-SA (16 mol eq), ST3Gal3 &
ST3Gal1

a-d, i-m, p-u, aa (independently selected) = 0 or 1;
o, z = 0; n, e-h = 1;
v-y (independently selected) = 0 or 1; R = PEG.

FIG. 36C

126/498

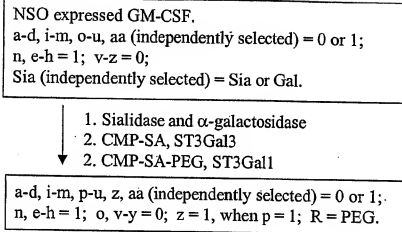


FIG. 36D

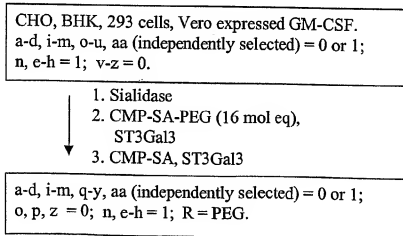


FIG. 36E

127/498

CHO, BHK, 293 cells, Vero expressed GM-CSF.
a-d, i-m, o-u, aa (independently selected) = 0 or 1;
n, e-h = 1; v-z = 0.



1. CMP-SA-levulinate, ST3Gal3,
buffer, salt
2. H₄N₂-PEG

a-d, i-m, o-y, aa (independently selected) = 0 or 1;
z = 0; n, e-h = 1; R = PEG.

FIG. 36F

CHO, BHK, 293 cells, Vero expressed GMCSF.
a-d, i-m, o-u, aa (independently selected) = 0 or 1;
n, e-h = 1; v-z = 0.

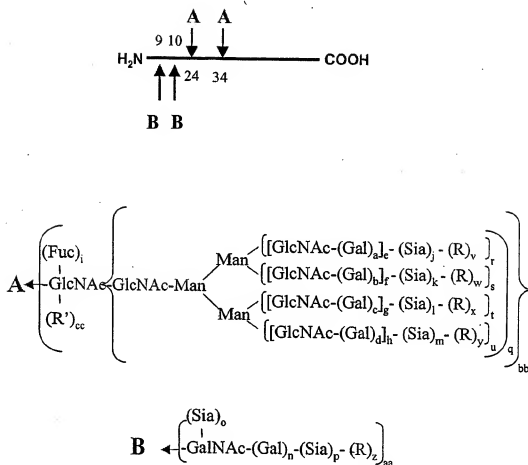


1. CMP-SA, α 2,8-ST

a-d, i, o-u, aa (independently selected) = 0 or 1;
n, e-h = 1; j-m (independently selected) = 0-20;
v-z (independently selected) = 0.

FIG. 36G

128/498



a-d, i, n-u, aa, bb, cc (independently selected) = 0 or 1.

c-h (independently selected) = 0 to 6.

j-m (independently selected) = 0 to 100.

v-y = 0; R = modifying group, mannose, oligo-mannose.

R' = H, glycosyl residue, modifying group, glycoconjugate.

FIG. 36H

129/498

Insect cell expressed GM-CSF.

a-d, f, h, j-m, o, p, s, u, v-z = 0;

c, g, i, n, q, r, t, aa (independently selected) = 0 or 1.

1. GNT's 1,2,4,5, UDP-GlcNAc
2. Galactosyltransferase, UDP-Gal-PEG

a-i, n, q-u (independently selected) = 0 or 1;

j-m = 0; v-y (independently selected) = 1,

when e-h (independently selected) is 1;

R = PEG.

FIG. 36I

Yeast expressed GM-CSF.

a-p, z, cc = 0;

q-y, aa (independently selected) = 0 to 1;

bb = 1; R (branched or linear) = Man, oligomannose;

GalNAc = Man.

1. Endoglycanase
2. mannosidase (if aa = 1).
3. Galactosyltransferase, UDP-Gal-PEG

a-p, r-z, aa, bb = 0;


q, cc (independently selected) = 0 or 1;

R' = -Gal-PEG.

FIG. 36J

130/498

CHO, BHK, 293 cells, Vero expressed GM-CSF.
a--m, o-u, aa, bb (independently selected) = 0 or 1;
n, v-z, cc = 0.

- 
1. sialidase
 2. CMP-SA, ST3Gal3
 2. CMP-SA-linker-SA-CMP, ST3Gal1
 3. ST3Gal3, transferrin

a--m, p-u, z, aa (independently selected) = 0 or 1;
o, v-y, cc = 0; bb, n = 1; R = transferrin.

FIG. 36K

132/498

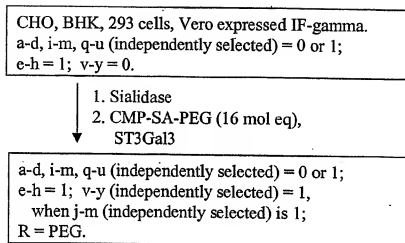


FIG. 37B

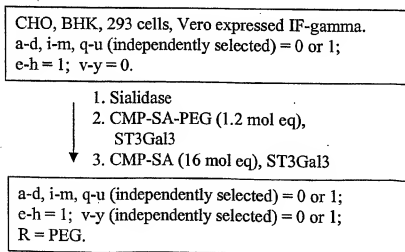


FIG. 37C

133/498

NSO expressed Interferon gamma.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0;
Sia (independently selected) = Sia or Gal.

- ↓
1. Sialidase and α -galactosidase
 2. α -Galactosyltransferase, UDP-Gal
 - ▼ 3. CMP-SA-PEG, ST3Gal3

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y (independently selected) = 1,
when j-m (independently selected) is 1;
R = PEG.

FIG. 37D

CHO, BHK, 293 cells, Vero expressed
Interferon gamma.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. Sialidase
 2. CMP-SA-PEG (16 mol eq),
ST3Gal3
 - ▼ 3. CMP-SA, ST3Gal3

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y (independently selected) = 0 or 1;
R = PEG.

FIG. 37E

134/498

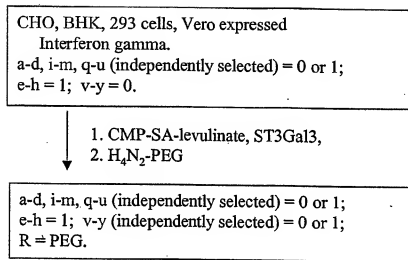


FIG. 37F

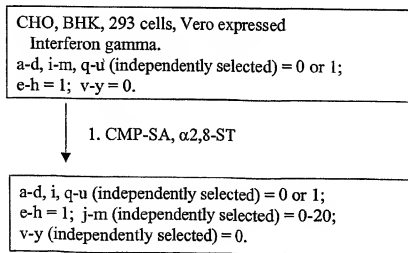
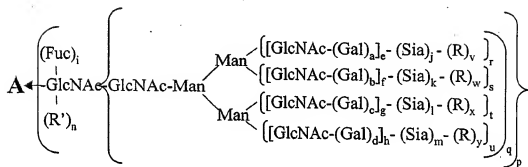
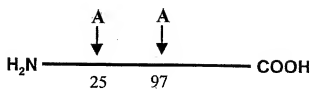


FIG. 37G

135/498



a-d, i, n, p-u (independently selected) = 0 or 1.

e-h (independently selected) = 0 to 6.

j-m (independently selected) = 0 to 100.

v-y = 0;

R = modifying group, mannose, oligo-mannose;

R' = H, glycosyl residue, modifying group,
glycoconjugate.

FIG. 37H

136/498

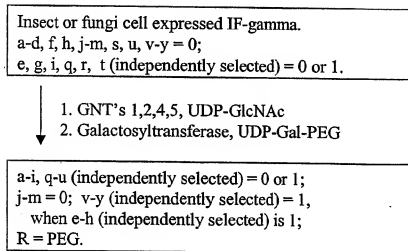


FIG. 37I

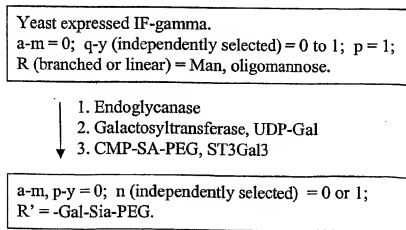


FIG. 37J

137/498

CHO, BHK, 293 cells, Vero expressed IF-gamma.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. CMP-SA-linker-Gal-UDP, ST3Gal3
 2. Galactosyltransferase, transferrin treated with endoglycanase.

a-m, q-u (independently selected) = 0 or 1;
p = 1; n = 0;
v-y (independently selected) = 0 or 1;
R = linker-transferrin.

FIG. 37K

CHO, BHK, 293 cells, Vero expressed
Interferon gamma.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h, p = 1; n, v-y = 0.

- ↓
1. CMP-SA-PEG,
ST3Gal3

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h, p = 1;
n, v-y (independently selected) = 0 or 1;
R = PEG.

FIG. 37L

138/498

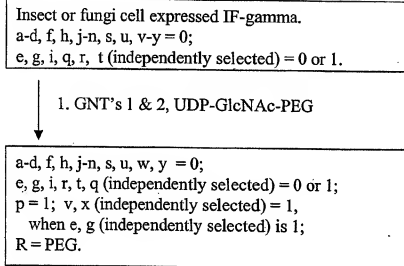


FIG. 37M

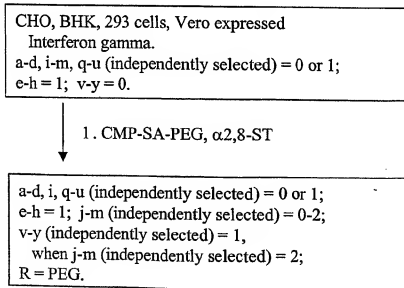


FIG. 37N

140/498

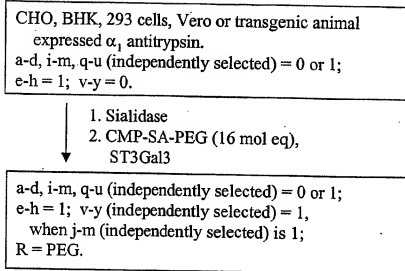


FIG. 38B

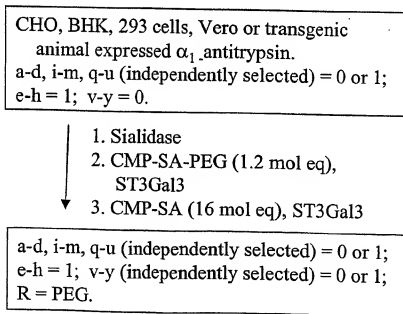


FIG. 38C

141/498

CHO, BHK, 293 cells, Vero or transgenic animal
expressed alpha-1 antitrypsin.

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. Sialidase
 2. CMP-SA-PEG (16 mol eq),
ST3Gal3
 3. CMP-SA, ST3Gal3

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y (independently selected) = 0 or 1;
R = PEG.

FIG. 38D

CHO, BHK, 293 cells, Vero or transgenic animal
expressed α_1 -antitrypsin.

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. CMP-SA-levulinate, ST3Gal3,
buffer, salt
 2. H_4N_2 -PEG

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y (independently selected) = 0 or 1;
R = PEG.

FIG. 38E

142/498

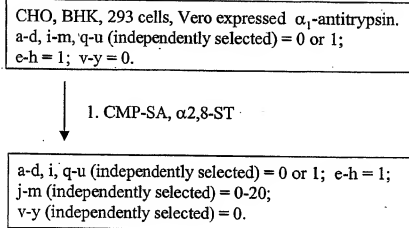
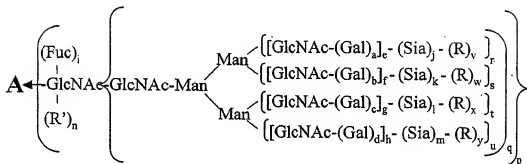
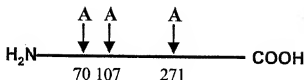


FIG. 38F

143/498



a-d, i, n, p-u (independently selected) = 0 or 1.

e-h (independently selected) = 0 to 6.

j-m (independently selected) = 0 to 100.

v-y = 0;

R = modifying group, mannose, oligo-mannose;

R' = H, glycosyl residue, modifying group, glycoconjugate.

FIG. 38G

144/498

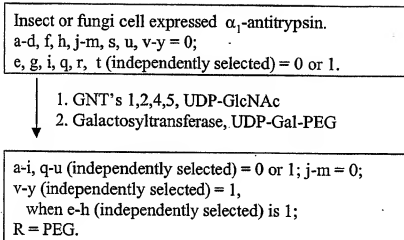


FIG. 38H

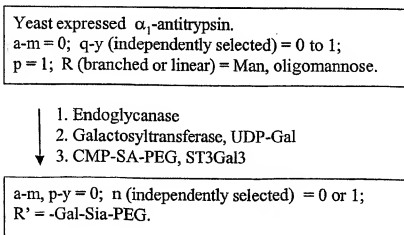


FIG. 38I

145/498

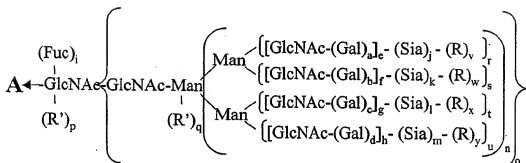
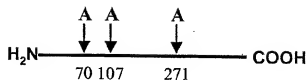
CHO, BHK, 293 cells, Vero expressed α_1 -antitrypsin.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. CMP-SA-linker-Gal-UDP,
ST3Gal3
 2. Galactosyltransferase, transferrin treated
with endoglycanase

a-m, q-u (independently selected) = 0 or 1;
p = 1; n = 0;
v-y (independently selected) = 0 or 1;
R = linker-transferrin.

FIG. 38J

146/498



a-d, i, n-u (independently selected) = 0 or 1.

e-h (independently selected) = 0 to 4.

j-m (independently selected) = 0 to 20.

R = polymer;

R', R'' (independently selected) = sugar, glycoconjugate.

FIG. 38K

147/498

Yeast expressed alpha-1 antitrypsin.

a-h, i-m, p, q = 0;

R (independently selected) = mannose, oligomannose, polymannose;

r-u, v-y (independently selected) = 0 or 1; n, o = 1.

↓ 1. endoglycanase

▼ 2. Galactosyltransferase, UDP-Gal-PEG

a-h, i-o, q, r-u, v-y = 0; p = 1.

R'' = Gal-PEG.

FIG. 38L

Plant expressed alpha-1 antitrypsin.

a-d, f, h, j-m, s, u, v-y = 0;

e, g, i, q, r, t (independently selected) = 0 or 1;

n = 1; R' = xylose

↓ 1. hexosaminidase,

2. alpha mannosidase and xylosidase

▼ 3. GlcNAc transferase, UDP-GlcNAc-PEG

a-d, f, h, j-n, s, u, v-y = 0;

e, g, i, r, t (independently selected) = 0;

q = 1; R' = GlcNAc-PEG.

FIG. 38M

148/498

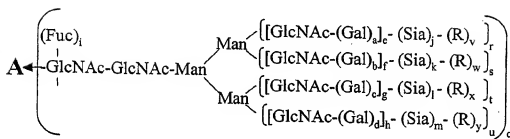
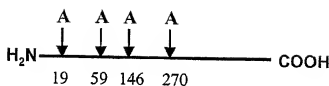
CHO, BHK, 293 cells, Vero, transgenic animal
expressed α_1 antitrypsin.
a-h, i-o, r-u (independently selected) = 0 or 1;
p, q, v-y = 0.

↓
1. CMP-SA-PEG,
ST3Gal3

a-h, i-o, r-u (independently selected) = 0 or 1;
p, q = 0; v-y (independently selected) = 0 or 1;
R = PEG.

FIG. 38N

149/498



a-d, i, q-u (independently selected) = 0 or 1.

e-h (independently selected) = 0 to 6.

j-m (independently selected) = 0 to 100.

v-y = 0; R = polymer.

FIG. 39A

150/498

CHO, BHK, 293 cells, Vero expressed Cerezyme
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. Sialidase
 2. CMP-SA-PEG (16 mol eq),
ST3Gal3

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y (independently selected) = 1,
when j-m (independently selected) is 1;
R = PEG.

FIG. 39B

CHO, BHK, 293 cells, Vero expressed Cerezyme.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. Sialidase
 2. CMP-SA-M-6-P (1.2 mol eq),
ST3Gal3
 3. CMP-SA (16 mol eq), ST3Gal3

a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y (independently selected) = 0 or 1;
R = mannose-6-phosphate

FIG. 39C

151/498

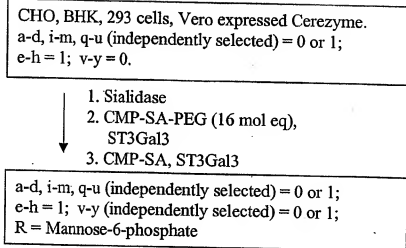


FIG. 39D

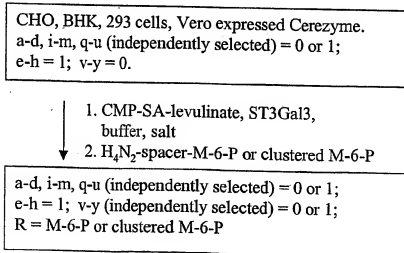


FIG. 39E

152/498

CHO, BHK, 293 cells, Vero expressed Cerezyme.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.



1. CMP-SA, α 2,8-ST

a-d, i, q-u (independently selected) = 0 or 1;
e-h = 1; j-m (independently selected) = 0-20;
v-y (independently selected) = 0.

FIG. 39F

154/498

Insect cell expressed Cerezyme.

a-d, f, h, j-m, s, u, v-y = 0;

e, g, i, q, r, t (independently selected) = 0 or 1.



1. GNT's 1,2,4,5, UDP-GlcNAc

2. Galactosyltransferase, UDP-Gal-PEG

a-i, q-u (independently selected) = 0 or 1;

j-m = 0;

v-y (independently selected) = 1,

when e-h (independently selected) is 1;

R = PEG.

FIG. 39H

Yeast expressed Cerezyme.

a-m = 0; q-y (independently selected) = 0 to 1;

p = 1; R (branched or linear) = Man, oligomannose.



1. Endoglycanase

2. Galactosyltransferase, UDP-Gal

3. CMP-SA-PEG, ST3Gal3

a-m, p-y = 0; n (independently selected) = 0 or 1;

R' = -Gal-Sia-PEG.

FIG. 39I

155/498

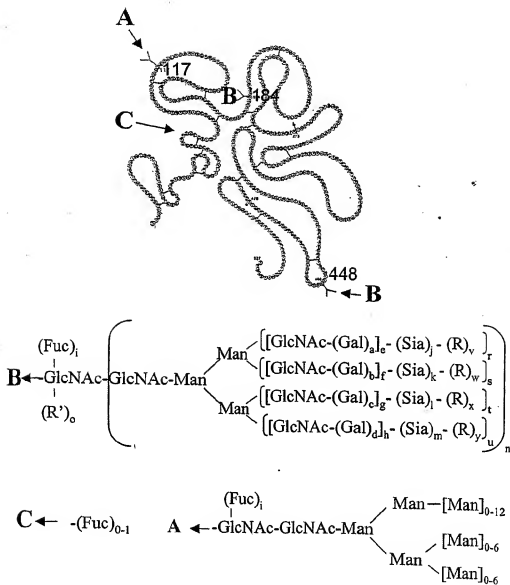
CHO, BHK, 293 cells, Vero expressed Cerezyme.
a-d, i-m, q-u (independently selected) = 0 or 1;
e-h = 1; v-y = 0.

- ↓
1. CMP-SA-linker-SA-CMP,
ST3Gal3
 2. ST3Gal3, desialylated transferrin.
 3. CMP-SA, ST3Gal3

a-m, q-u (independently selected) = 0 or 1;
p = 1; n = 0; v-y (independently selected) = 0 or 1;
R = linker-transferrin.

FIG. 39J

156/498



a-d, i, n-u (independently selected) = 0 or 1.
 e-h (independently selected) = 0 to 4.
 j-m (independently selected) = 0 to 20.
 R = polymer; R' = sugar, glycoconjugate.

FIG. 40A

157/498

CHO, BHK, 293 cells, Vero expressed tPA
 a-g, n = 1; h = 1 to 3;
 j-m, i, (independently selected) = 0 or 1;
 r-u (independently selected) = 0 to 1; o, v-y = 0.

1. Mannosidase(s), sialidase
2. GNT1,2 (4 and/or 5) UDP-GlcNAc
3. Gal transferase, UDP-Gal
- ▼ 4. CMP-SA-PEG, ST3Gal3

A = B; a-g, n = 1; h = 1 to 3;
 i, r-u (independently selected) = 0 or 1;
 o = 0; j-m, v-y (independently selected) = 0 or 1;
 R = PEG

FIG. 40B

Insect or fungi cell expressed tPA
 A = B; a-d, f, h, j-o, s, u, v-y = 0;
 e, g, i, n, r, t (independently selected) = 0 or 1.

1. GNT's 1&2, UDP-GlcNAc
2. Galactosyltransferase, UDP-Gal
- ▼ 3. CMP-SA-PEG, ST3Gal3

A = B; b, d, f, h, k, m, o, s, u, w, y = 0;
 a, c, e, g, i, r, t (independently selected) = 0 or 1;
 n = 1; j, l, v, x (independently selected) = 0 or 1;
 R = PEG.

FIG. 40C

158/498

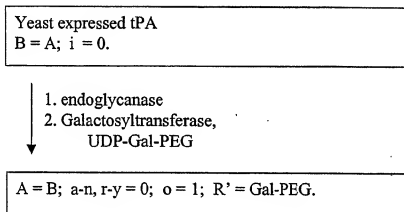


FIG. 40D

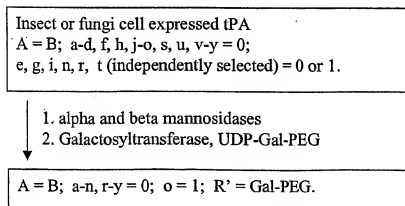


FIG. 40E

159/498

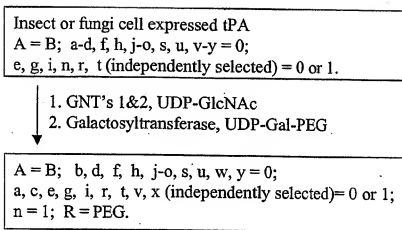


FIG. 40F

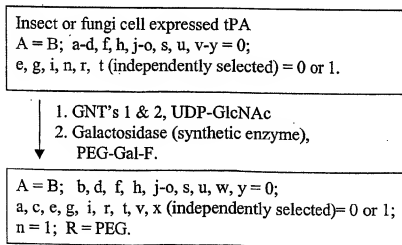


FIG. 40G